Evaluation of Experimental Antimalarial Drugs for Radical Curative Activity in the Rhesus Monkey

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OBJECTIVE : To evaluate the radical curative effectiveness of selected experimental drugs in rhesus monkeys (*Macaca mulatta*) infected with *Plasmodium cynomolgi* malaria.

BACKGROUND : This is a continuation of studies initiated by this Laboratory in 1974. A chronological report of the methodology and results are available in previous SEATO/AFRIMS Annual Reports (1, 2). These studies are conducted in association with the Department of Parasitology, Division of Experimental Therapeutics, Walter Reed Army Institute of Research.

METHODS : Rhesus monkeys were inoculated intravenously with sporozoites produced in *Anopheles dirus* mosquitoes.

A. dirus mosquitoes were fed on *P. cynomolgi* infected monkeys. This feeding was conducted during the second or third rise in parasitemia and when both male and female gametocytes were present as evidenced by a blood smear. On post-feeding day 14, the sporozoites were harvested from the salivary glands of the infected mosquitoes and diluted in a saline-normal monkey serum solution (1:1) to a concentration of 5-20 x 10^5 sporozoites per ml. Preselected, malaria-negative rhesus monkeys were immediately inoculated intravenously with one ml. of the sporozoite solution.

Each monkey was monitored by blood smears daily, beginning on day 7 post-treatment for the development of a parasitemia. When the parasitemia reached 5-25 x 10^3 parasites per cmm, test drugs were administered daily for seven days at a predetermined dosage level, based on a mg. of drug/kg. of body weight. To permit evaluation of drug activity against tissue parasitic forms independently of blood schizonticidal activity, chloroquine phosphate was administered simultaneously with each test drug at 5 mg/kg. body weight/day for seven days.

Following administration of the test drug, malaria parasitemia was monitored by examination of Giemsa stained blood smears daily for twelve days and on Monday, Wednesday and Friday thereafter. Prior to 1 March 1978, monkeys

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which converted to a negative parasitemia and remained so through post-treatment day 20 were splenectomized and monitored an additional 33 days. Those that remained free of malaria parasites during this period were considered cured. After 1 March 1978, monkeys which converted to a negative parasitemia were monitored for 80 days post-treatment with no splenectomy. Those remaining negative during this period were considered cured. Those monkeys which either failed to convert to negative parasitemias or which did convert to a negative status initially but subsequently became positive again in under 20 days post-treatment were considered not cured. These monkeys are terminated on the particular drug study when this occurs, however, when their parasitemias reach an acceptable level (approximately 5,000/cmm) they are placed on another, different drug. In this manner, it is possible for one monkey to be used to test several drugs provided they "break" with a parasitemia before post treatment day 20.

RESULTS : A total of 32 experimental drugs were evaluated: results are summarized in Tables 1 & 2. This reduced number of drugs tested over previous years is a direct result of the world-wide shortage of rhesus monkeys available for medical research.

REFERENCES

1. Brown, J.L., et.al., Annual Progress Report, SEATO Medical Research Laboratory, April 1975 - March 1976, pp. 133-135.

2. Brown, J.L., et.al., Annual Progress Report, AFRIMS, April 1976 - September 1977, pp. 155-158.

Table 1. Summary of Completed Sporozoite Induced Tests in Rhesus Monkeys

Type of Compound	WPAIR Drug Number	Minimum Curative Dose* (mg/kg/day)
8 - Aminoquinoline	234073	** NC(10.0)
	237808	10.0
	238403	NC(10.0)
	238599	NC(3.16)
	238887	NC(3.16)
	238872	NC(10.0)
	239124	NC(10.0)
	239378	NC(3.16)
	240381	NC(3.16)
	241319	NC(10.0)
	234075	NC(0.1)
Acridines	235660	NC(10.0)
	233602	NC(10.0)
	239373	NC(3.16)
Quinolines	229238	1.0
Miscellaneous	232651	NC(10.0)
	235963	NC(10.0)
	237375	NC(3.16)
	240595	NC(10.0)

* Administered orally with 5.0 mg/kg/day of chloroquine phosphate.

** Not Curative - The compound did not cure at the maximum dose tested or tolerated. The maximum dose is indicated in parentheses.

Table 2. Summary of Drugs Currently Undergoing Sporozoite Induced Tests in Rhesus Monkeys

Type of Compound	WRAIR Drug Number	Dosage Level Currently Being Tested (mg/kg/day)
8 - Aminoquinoline	237807	10.0, 1.0, 3.16, 0.316
	238605	1.0, 0.316, 0.1
	238608	10.0, 3.16, 1.0
	238850	1.0, 3.16
	239372	1.0, 0.316
	241320	1.0
	242047	1.0
	242471	1.0
	242511	1.0, 0.316, 0.1
Quinoline	3863-D-0	1.0
Miscellaneous	230190	1.0
	234737	1.0
	237797	10.0, 1.0, 0.316